TRANSPARENCY AND MOTTLING, A CASE OF MENDELIAN INHERITANCE IN THE GOLDFISH *CARASSIUS AURATUS**

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INTRODUCTION

In the spring of 1924 I made a preliminary study of the heredity of goldfish by crossing various breeds of goldfish and mating domesticated breeds of goldfish with the wild goldfish. During the progress of this preliminary study I noticed that the inheritance of one of the characters under investigation was rather simple.

This character was called "transparent scale" in my previous paper (CHEN 1925). The goldfishes of this breed have only a few normal scales, the remainder of the body being apparently naked. The apparently naked part of the body is really covered by scales which, on account of the lack of a layer of reflecting tissue on their inner side, are as transparent as glass. This breed of goldfish is called "shubunkin" in Japan (MATRUBARA 1908) and is known as "calico" in the United States (INNES 1917).

Beginning in the spring of 1925 I started more extensive experiments to investigate the mode of inheritance of this character, "transparent scale"or "calico." In the summer of 1925 I obtained enough evidence to prove that the inheritance of this character is Mendelian. A preliminary report of this discovery had been made (CHEN 1926). The present paper is the final report of this investigation.

* The GALTON AND MENDEL MEMORIAL FUND contributes the accompanying colored illustrations.

MATERIAL AND METHODS

The materials of this investigation were the transparent scaled fancy goldfishes, the normal scaled fancy gold..shes, and the normal scaled wild goldfishes. The fancy goldfishes were obtained from a local goldfish breeder, the wild ones being obtained from the food-fish market of Nanking.

The fishes were bred and grown in water reservoirs made of burned clay. These reservoirs were made in the shape of a huge cup with the upper diameter about 38 inches, lower diameter about 19 inches, and a depth of about 32 inches. Such a reservoir is large enough to contain about eighty young fishes to a size capable of reproduction in the next breeding season.

Goldfish take one year for a generation. The breeding season of the goldfish at Nanking begins in the middle of April and ends in the middle of June, lasting about two months. During this period a single male fish may mate four or five times and a female fish two or three times with an interval of about ten days between successive matings.

I tried to make the different crosses of goldfish by means of artificial fertilization. These attempts failed. Hence all the various crosses in my experiments were made by the ordinary method of natural fertilization as employed by the common goldfish breeders except with some necessary precautions.

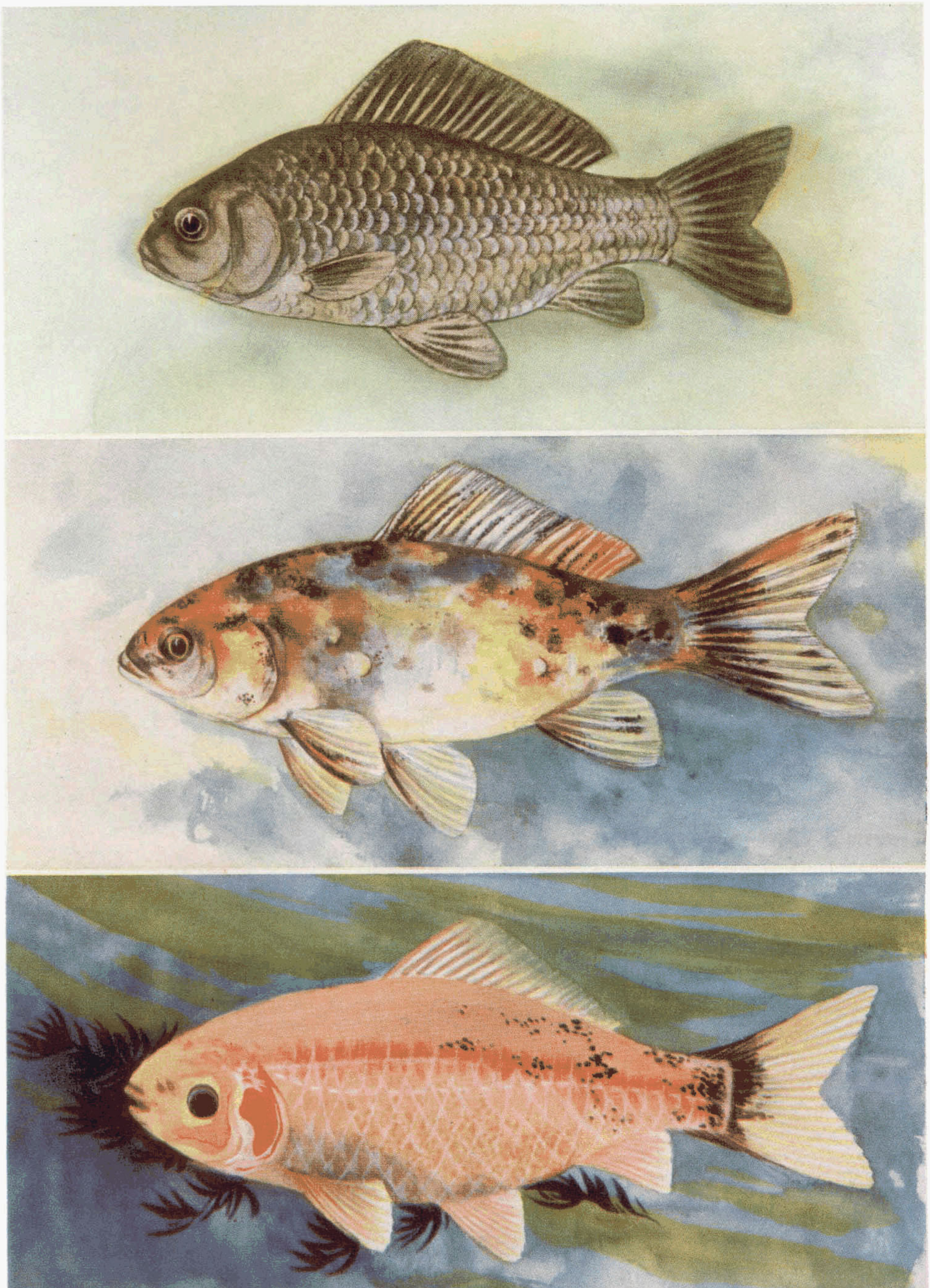
At the beginning of the breeding season I separated the female and male fishes into different reservoirs. The male fish is distinguished from the female by the presence of numerous small white tubercles, called the "pearl organs," along the front rays of the pectoral fins and on the external side of the two opercula. This separation of the females from the males was needed in order to avoid the loss of the eggs through the free matings in a reservoir as soon as the breeding season began and thus to insure the females to have eggs to lay when these were required for the experiments.

In mating goldfish the practice of the common breeders is to mate two or three female goldfishes with three or four males. Since such a practice would introduce some errors into the experimemts, I always mated one female with one male in a reservoir.

During mating the eggs of the gold sh were deposited on a bunch on water plant (Myriophyllum) which had been introduced into every mating reservoir. The eggs hatch from three days to a week after fertilization depending upon the temperature of the water in the reservoir.

LEGEND FOR PLATE 1

FIGURE 1.—Normal scaled fish.
FIGURE 2.—Heterozygous mottled fish.
FIGURE 3.—Homozygous transparent fish.



About two days after hatching, when the young fish were just able to swim freely, they were fed with finely pulverized yolk of boiled chick eggs. About three days later I began to feed the young fish with living cyclops and about two weeks after hatching the young fishes were fed with Daphnia. The goldfishes grew very rapidly on this diet.

About forty days after hatching the young fish had grown to about 25 mm in body length, being a size large enough to be examined with the naked eye. At this time the character of transparent scale had developed to such an extent that the fish with this character might be distinguished from the normal scaled fishes. The data concerning the offspring of the matings were mostly taken at this time. Sometimes the progeny of a single mating amount to about seven or eight hundred.

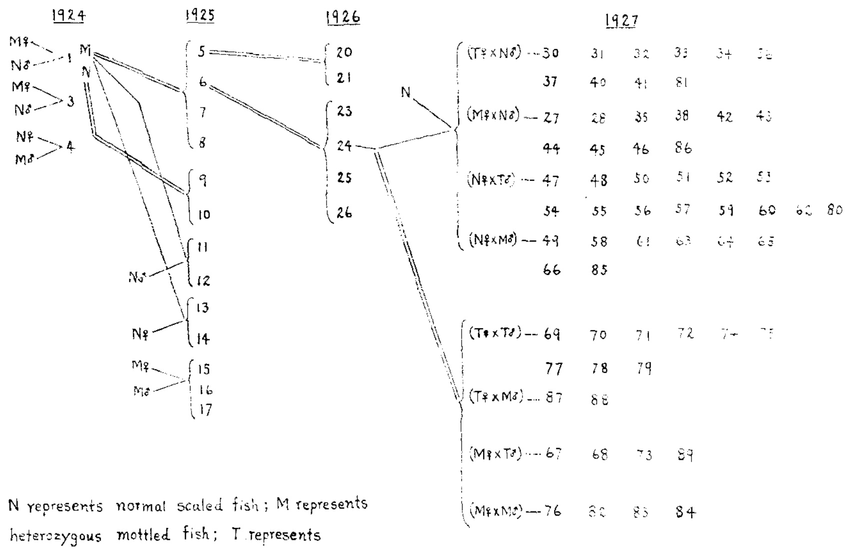
Frequently the young offspring were discarded after being examined, classified, and recorded. Sometimes these fishes were reserved for further examination. The young fishes at this time, about forty days after hatching, varied greatly in size. Goldfishes are cannibals and if the small fishes remain with the larger ones, the latter will devour the former. In a heterogeneous group of genotypically different classes of fish the quick growing will soon devour the slow growing and the former will survive while the latter perish. In order to avoid the introduction of this source of error into my experiments, I always separated the offspring of a single mating, at the age of about forty days after hatching, into two groups according to the size of the young fishes and reared these two groups separately in different reservoirs. Thus the slow-growing class had as good a chance to survive as the quick-growing class.

NARRATIVE OF THE EXPERIMENTS

1924. The parents and the F_1—In the spring of 1924 I made the following crosses (figure 1):

1. Transparent scaled fancy goldfish ♀ Normal scaled wild goldfish ♂
2. Normal scaled wild goldfish ♀ Transparent scaled fancy goldfish ♂
3. Transparent scaled fancy goldfish ♀ Normal scaled fancy goldfish ♂
4. Normal scaled fancy goldfish ♀ Transparent scaled fancy goldfish ♂

With the exception of the mating number 2, all of the matings produced offspring. The F_1 fishes of all the fertile matings were unexpectedly of two classes. One class of the offspring had transparent scales and mottled condition while the other class had normal scales. The normal scaled offspring of family 1 were exactly like the wild goldfish (plate 1). The transparent-scaled offspring of the same family were similar to the wild goldfish in every respect except with transparent scales and the mottled pattern (plate 1, figure 2). The latter class of fish with a long body and unpaired fins resembles closely a Japanese breed of goldfish called "shubunkin" but is an entirely new form to the goldfish fanciers in China. I found both females and males among the transparently scaled and the normally scaled classes.



N represents normal scaled fish; M represents heterozygous mottled fish; T represents homozygous transparent fish.

The two classes of offspring were of approximately equal numbers as shown by the data in table 5. This result was interpreted by assuming that the transparent scaled parents of the families 1, 3, and 4 were all heterozygous with respect to a pair of allelomorphic factors one of which when homozygous was responsible for the production of the transparent scales while the other factor, when homozygous, would cause the production of the normal scales. All the normal scaled parents were homozygous for normal scales. The gene for the production of the transparent scales is accordingly dominant to the gene for the production of the normal scales. Hence the transparent scales and the mottled pattern appeared on the body of the heterozygous fish and the offspring of the three families were of the two classes, heterozygous mottled fish and homozygous normal scaled fish.

1925. The F_2 — From the offspring of family 1 I made four matings between the transparent scaled females and the similarly scaled males. The offspring of these matings were, as expected, of two classes, transparent scaled and normal scaled. The number of individuals in these two classes and their ratios are presented in table 3, families 5, 6, 7, and 8.

The data in table 3 follow closely the typical Mendelian ratio of the F_2 generation, three to one. The assumption that the original parents of 1924 were heterozygous and all the transparent scaled offspring were also heterozygous was thus confirmed.

In addition to the four matings mentioned above, I had also made four backcrosses between the transparent scaled offspring of family 1 and the normal scaled fancy goldfishes. The results of these matings are presented in table 5, families 11, 12, 13 and 14. The data of these back crosses also prove that the transparent scaled offspring were heterozygous as expected.

The matings between the normal scaled females and males of the offspring of family 1 had also been made. One of these matings produced 761 normal scaled offspring (family 9). Another mating produced 248 normal scaled offspring (family 10). Neither of these matings produced a single transparent scaled fish. This result is in accord with the assumption that the original normal scaled parents and the normal scaled offspring were homozygous and that the normal scaled condition is recessive.

1926 The F_3 generation—On account of a necessary absence from Nanking during the breeding season of 1926 I did not have time to make more extensive experiments than nine matings between the transparently scaled females and males of the F_2 generation.

Only six out of the nine matings produced offspring forming the families 20, 21, 23, 24, 25, and 26. Families 20, 21, 23, 25, and 26 consisted of transparent scaled and normal scaled offspring in the typical Mendelian ratio of three to one (table 3). Family 24, however, consisted of 346 transparent scaled fishes together with only one normal scaled fish. The exceptional normal scaled fish was thought to have been accidently introduced into this family from some other source due to the carelessness of the aquarium keeper during my absence from Nanking.

1927 The F_4 generation.—In the spring of 1927 I took twenty females and twenty males from the offspring of family 24 and backcrossed them with normal scaled goldfishes. I obtained the following results:

Nine transparent scaled females and thirteen transparent scaled males in mating with the normal scaled goldfish produced only transparent scaled offsprings. These twenty-two transparent scaled fishes were thus proved to be homozygous with respect to the gene for the transparent scale.

Nine transparent scaled females and seven transparent scaled males in mating with the normal scaled goldfishes produced offspring of two kinds in approximately equal numbers. These sixteen transparent scaled fishes were thus proved to be heterozygous, with one gene for transparent scale and one gene for normal scale.

Two transparent scaled females in the backcross with normal scaled goldfishes failed to produce offspring.

By the above mentioned breeding work I proved twenty-two transparent scaled fishes to be homozygous and sixteen transparent scaled fishes to be heterozygous. These two classes of fishes were examined in detail. I found that they have distinguishing features which although difficult to define in terms of words, are, on the whole, easily recognized as soon as it has once been noted. Figure 2 of plate 1 shows a heterozygous transparent scaled fish. Figure 3 of the same plate shows a homozygous transparent scaled fish. Detailed descriptions will be made later.

After I ascertained the genotypes of the thirty-eight transparent scaled fishes I made the following matings between the females and the males of these thirty-eight fishes near the end of the breeding season:

1. Nine matings between the females and the males of the homozygous transparent scaled fishes.
2. Two matings between the homozygous transparent scaled females and the heterozygous males.
3. Four matings between the heterozygous females and the homozygous transparent scaled males.
4. Four matings between the females and the males of the heterozygous fishes.

The above matings were made for the double purpose of testing whether the earlier assumptions were true and to see whether the ratios of the number of homozygous transparent scaled fishes to that of the heterozygous fishes were approximate to the expected ratios.

The results of these matings are presented in tables 1, 4, and 6. The data in these tables show that observed ratios are in close accordance with expectation.

TERMINOLOGY AND SYMBOLS

At the beginning of the present investigation I could distinguish only one kind of transparent scaled goldfish. This breed of goldfish is called "shubunkin" in Japan and "calico" in the United States.

After four years of experiments I found that the transparent scaled fishes could be separated into two classes, one of which is nearly completely transparent while the other class always has a few normal scales and the characteristic mottled pattern. The former class is homozygous and the latter class heterozygous.

Since the homozygous transparent scaled fishes are phenotypically distinguishable from the heterozygous transparent scaled fishes, hereafter I shall call the fishes of the former class "transparent fish" and the fishes of the latter class "mottled fish."

The distinction between heterozygous mottled and normal fish is very sharp but the distinction between heterozygous and homozygous transparent fish is not so sharp and they even overlap when the fishes are young. According to the terminology of T. H. MORGAN (1919) a character is said to be recessive if the individuals with this character are sharply separable from the heterozygous individuals; a character is said to be dominant if the individuals with this character are overlapping with the heterozygous individuals. In this sense we may say that the "transparency" is a dominant character while the normal condition is recessive.

According to the methodology of T. H. MORGAN the symbol of the gene causing a dominant mutant character, like the bar eye of Drosophila, is a capital letter primed while the symbol to represent the gene causing the normal recessive character is the capital letter alone. Adopting this method of MORGAN I shall use the letter T to represent the gene causing the development of normal scales and the symbol T' to represent the gene causing the development of transparent scales.

DATA FROM BREEDING EXPERIMENTS

Normal scaled fishes breed true.—During the period of 1924 to 1927 I made numerous matings between the females and males of the normal scaled fancy goldfishes as well as the wild goldfishes. With the exception of a very few instances to be cited in a later section of this paper the offspring of all these matings consist of only normal scaled fishes. Furthermore I made two matings (families 9 and 10) of normal scaled fishes produced from a cross (family 1) between a transparent scaled fish and a normal scaled fish. One of the matings (family 9) produced 761 normal scaled fishes. The other mating (family 10) produced 248 normal scaled fishes. Both of these two matings did not produce a single fish with transparent scales.

Transparent fishes breed true.—In the spring of 1927 I made nine matings between the female and male transparent fishes. These nine matings produced many thousands of offspring, all of them were transparent fishes. The data are in table 1.

TABLE 1
Families which illustrate the true-breeding of transparency.

PEDIGREE NUMBERS			NUMBER OF OFFSPRING, ALL TRANSPARENT
Parents		Family	
♀	♂		
24(B5)*	24(C5)	69	81
(B6)	(C6)	70	183
(B7)	(C7)	71	41
(B8)	(C8)	72	122
(B10)	(C10)	74	150+
(B11)	(C11)	75	150+
(B4)	(C1)	77	100+
(B5)	(C2)	78	100+
(B6)	(C4)	79	100+
Total			1027+

* The number outside of bracket represents family number; those within the brackets represent individual numbers.

Transparent fish crossed with normal fish produce mottled fish.—In the spring of 1927 I made ten crosses between transparent females and normal males and fourteen crosses between normal females and transparent males. All these matings produced only fishes with the characteristic mottled pattern. The data of these matings are in table 2.

TABLE 2
Families which show the mottled heterozygous type in the F_1 of crosses between transparent and normal.

PEDIGREE NUMBERS		NUMBER OF OFFSPRING ALL MOTTLED	PEDIGREE NUMBERS		NUMBER OF OFFSPRING, ALL MOTTLED
♀ Parent*	Family		♂ Parent**	Family	
24(B4)	30	150+	24(C1)	47	350+
(B5)	31	174	(C2)	48	100+
(B6)	32	150+	(C4)	50	300+
(B7)	33	14	(C5)	51	300+
(B8)	34	100+	(C6)	52	200+
(B10)	36	80+	(C7)	53	100+
(B11)	37	150+	(C8)	54	100+
(B14)	40	100+	(C9)	55	300+
(B15)	41	14	(C10)	56	100+
(B10)	81	100+	(C11)	57	200+
			(C13)	59	200+
Total		1032+	(C14)	60	200+
			(C16)	62	200+
			(C5)	80	100+
			Total		2750+

* ♂ parents were normal scaled.
** ♀ parents were normal scaled.

Mottled fish never breed true, but always produce some normal fish and some transparent fish.—In the spring of 1925 I made seven matings between female and male mottled fishes. In the spring of 1926 I made five similar matings. The data of these matings are in table 3. These data show that among the offspring there were always produced some normal fishes. The ratios of the transparent and mottled fishes to the normal scaled fishes were all approximate to 3 to 1. The deviation of the observed ratios

TABLE 3

Segregations following mating mottled females with mottled males.

FAMILY NUMBER	TRANSPARENT AND MOTTLED OFFSPRING		NORMAL SCALED OFFSPRING		TOTAL NUMBER OF OFFSPRING
	Number	Ratio	Number	Ratio	
5	532	2.86	212	1.14	744
6	856	3.01	280	.99	1136
7	508	2.87	201	1.14	709
8	744	2.87	291	1.12	1035
15	482	3.08	143	.92	625
17	119	3.17	31	.83	150
20	236	2.84	96	1.16	332
21	181	3.07	55	.93	236
23	152	2.81	64	1.19	216
25	375	2.98	128	1.02	503
26	301	3.17	79	.83	380
Totals	4486	2.958	1580	1.042	6066
P.E.		±0.015		±0.015	

TABLE 4

Segregations following mating mottled females with mottled males.

PEDIGREE NUMBERS			NORMAL SCALED OFFSPRINGS		MOTTLED OFFSPRING		TRANSPARENT OFFSPRING		TOTAL NUMBER OF OFF-SPRING
Parents ♀	♂	Family	Number	Ratio	Number	Ratio	Number	Ratio	
24(B12)	24(C12)	76	71	.83	170	1.98	102	1.19	343
(B2)	(C3)	82	110	.82	274	2.18	120	.95	504
(B9)	(C12)	83	82	1.27	117	1.80	60	.93	259
(B12)	(C15)	84	82	1.00	168	2.05	78	.95	328
Totals			345	0.96	729	2.03	360	1.00	1434
P.E.				±0.03		±0.036		±0.03	

from this expectation is 0.042 while the probable error of the expectation is 0.015. Since the deviation is less than three times the probable error the fitness of the observed ratio to the expectation is reasonably close.

In the spring of 1927 I made four additional matings between female and male mottled fishes. The results of these matings are in table 4 which show that among the offspring there were always about one fourth of transparent fish, one half of mottled fish, and one fourth of normal fish. The observed ratio is very close to the expectation. In the case of the total normal to the total offspring the observation is in perfect agreement with the expectation. The deviations of the other two ratios are only about once times the probable error.

Mottled fish crossed with normal fish.—I made fourteen matings of mottled females crossed with normal males and eleven matings of normal females crossed with mottled males during the breeding seasons of 1924 to 1927. Among these 25 matings I obtained quantitative data in eleven of them. The data are presented in table 5. I had also made a rough examination of the results of the 14 remaining matings and found them to consist of about one half of mottled fishes and one half of normal scaled fishes.

TABLE 5

Mottled heterozygotes backcrossed to the normal.

FAMILY NUMBERS	TYPE OF CROSS	MOTTLED OFFSPRING		NORMAL SCALED OFFSPRING		TOTAL NUMBER OF OFFSPRING
		Number	Ratio	Number	Ratio	
1	M♀×N♂*	156	1.08	133	.92	289
3	M♀×N♂	33	1.12	26	.88	59
11	M♀×N♂	654	1.05	592	.95	1246
12	M♀×N♂	557	1.04	511	.96	1068
43	M♀×N♂	127	1.06	112	.94	239
86	M♀×N♂	23	.79	35	1.21	58
4	N♀×M♂	120	1.05	111	.95	231
13	N♀×M♂	146	.92	171	1.08	317
14	N♀×M♂	573	.99	580	1.01	1153
64	N♀×M♂	101	1.06	90	.94	191
85	N♀×M♂	62	1.02	59	.98	121
Totals		2552	1.03	2420	.97	4972
P.E.			±0.01		±0.01	

* N represents normal scaled fish; M represents heterozygous mottled fish.

The data in table 5 show that all the matings produced about one half of mottled fish and one half of normal fish. The observed ratio fits very closely to the expectation since the probable error of the expectation is ± 0.01 and the deviation of the observed ratio from the expectation is 0.03, being only three times the probable error.

Mottled fish crossed with transparent fish.—In the spring of 1927 I made two matings between transparent female and mottled male fishes and four matings between mottled female and transparent male fishes. The results of these matings are in table 6 which show that all these matings produced

TABLE 6

Mottled heterozygotes backcrossed to the transparent.

PEDIGREE NUMBERS			TYPE OF CROSS	MOTTLED OFFSPRING		TRANSPARENT OFFSPRING		TOTAL NUMBER OF OFF-SPRING
Parents ♀	Parents ♂	Family		Number	Ratio	Number	Ratio	
24(B7)	24(C18)	87	T♀×M♂*	123	1.00	124	1.00	247
(B11)	(C19)	88	T♀×M♂	145	1.07	127	.93	272
(B1)	(C1)	67	M♀×T♂	24	.89	30	1.11	54
(B2)	(C2)	68	M♀×T♂	28	1.06	25	.94	53
(B19)	(C9)	89	M♀×T♂	138	.85	188	1.15	326
Totals				458	.96	494	1.04	952
P.E.					± 0.02		± 0.02	

* T represents homozygous transparent fish.

about one half of mottled fish and one half of transparent fish. The observed ratio fits very closely to the expectation since the deviation is only two times the probable error in the total offspring.

Besides the above observations families 24 and 73 had been roughly examined and found to consist of about one half of transparent fishes and one half of mottled fishes.

The character is not sex-linked.—During the progress of the breeding work I always paid attention to the problem of whether the character is sex-linked or not. Since the secondary sexual character, that is, pearl penis organ, of the male fish, is not visible before the fish is grown to considerable size I often dissected many young fishes to ascertain their sex. In all the various matings which I made I found that there were always female and male offspring in approximately equal numbers. Based upon all the facts which I observed in the breeding experiments I conclude that this character is not sex-linked no matter whether the inheritance of sex in fish be XY type, WZ type, or the modified XY type with genes on the Y chromosome as well as on the X chromosome as found in the case of *Aplocheilus latipes* by AIDA (1921).

THE MANIFOLD EFFECTS

I have already mentioned that by means of the progeny tests I had definitely found 22 transparent scaled fishes to be homozygous and 16 transparent scaled fishes to be heterozygous. With the two classes of transparent scaled fishes and the normal scaled fishes at my disposal I made a detailed examination of the various effects of the different combinations of the allelomorphic genes T and T'. This study is extended and confirmed by the examination of the offspring produced by the matings of the parents whose genotypic constitutions are definitely known.

Homozygous Normal.—With the presence of the gene T in duplex condition the fish has one or several layers of reflecting tissue covering the entire body including the head, trunk, and iris and excluding the pupils and the fins, being more abundant on the ventral surface of the body. The fish has also two kinds of chromatophores, that is, the black melanophores and the yellow xanthophores, distributed on the surface of the body and the fins and usually more abundant on the dorsal surface than on the lateral surface while on the ventral surface the chromatophores are generally absent.

With the exception of the breeds "brown" and "blue" the fishes with the gene T in duplex condition always have similar body color until about ten weeks after hatching, beginning in July, when the various brilliant colors of goldfish begin to appear.

Homozygous Transparent.—When the gene T' is in the duplex condition the fish has an appearance (plate 1, figure 3) entirely different from that of normal scaled fishes. The gene reduces the reflecting tissue so that it is either entirely, or nearly entirely absent on the body of the fish thus showing the red blood in the gills, the black pigment in the eyeballs, the white testis, the yellow eggs, and the black intestine visible to the eye without dissection. The melanophores are also greatly reduced to a black dotted area near the junction between the caudal peduncle and the caudal fin while on the other parts of the body the melanophores are either absent or, when present, form isolated dots very few in number. The presence of T' in duplex condition causes usually the complete loss of xanthophores. Occasionally xanthophores are present and form a yellow spot on one side of the trunk.

The expression of the genes varies in different individuals. Among the twenty-two homozygous transparent fishes the effects on the reflecting tissue varied as follows:

Five fishes were completely transparent, that is, without reflecting tissue altogether.

Fourteen fishes had a very small area of reflecting tissue situated on the ventral or the ventro-lateral parts of the abdomen.

Seven fishes had some reflecting tissue on one or both of the irises.

Four fishes had some reflecting tissue on a part of an operculum or a whole operculum but never on both opercula.

Three fishes had one to three normal scales. Nineteen fishes had no normal scale.

The effects on the melanophores and the xanthophores also varies in different individuals. Generally the melanophores are restricted to the junction between the caudal peduncle and caudal fin while the xanthophores are entirely absent. But I had also frequently observed that the melanophores are present on the basal area of the dorsal fin, on the basal area of the anal fin, or on the top of the head, and the xanthophores are present on a small area of the surface of the body usually beneath the dorsal fin.

Heterozygous Mottled.—The expression of the effects of a gene T and a gene T' in combination is more complex than when the genotypic constitution is TT or $T'T'$. The head is sometimes completely covered with the reflecting tissue, sometimes transparent in a small portion of the skin, or sometimes lacking the reflecting tissue in an entire operculum or even in an area as large as about one half of the surface of the head.

In the heterozygous mottled fishes there are always some normal scales mixed with the transparent scales. The number of normal scales in the 16 heterozygous fishes which I studied in detail varied from 2 to 18, being most frequently from 2 to 10.

In the body wall of the trunk the inner layer of the reflecting tissue is either entirely present or reduced to about two-thirds of the whole surface. Hence the heterozygous fish is never completely transparent.

The melanophores, xanthophores, and the bluish prismatic color are always mixed in the characteristic mottle condition and there is a greater number of the colored spots on the dorsal and the dorsolateral parts of the body than on the ventral parts.

WHY TRANSPARENT FISHES ARE RARE

In reviewing the books on goldfish published in Japan, such as that of MATRUBARA (1908), or in the United States such as that of INNES (1917), I found no records of the existence of the transparent fish as distinguished from the mottled fish, "shubunkin," or "calico." In the Chinese books on goldfish I had also found no records of the transparent fish as a distinct breed. In my previous paper (CHEN 1925) I described such a fish but at that time I was not aware that such a fish might be different from the mottled fish in genotype and form a distinct breed. Altogether I mated nine transparent scaled fishes obtained from the local goldfish breeders at Nanking and found that all of them were heterozygous (families 1, 3, 4, 15, 16, 17). All these facts show that the transparent fish is rarely found in the goldfish aquarium of the ordinary goldfish breeders.

But in my breeding work I found that the proportion of the transparent fish in the F_4 generation is not less than one fourth. The transparent fish did not seem to have a poor viability as ordinary recessive breeds of animals or plants. I suggest the following three explanations to be the cause of the rarity of the transparent fish in the aquarium of the common goldfish breeders:

One explanation is that the heterozygous class being twice as numerous as the homozygous class has a greater chance to be obtained from the goldfish breeders. Another explanation is that the goldfish breeders often put five or six fishes into a breeding basin and frequently a normal scaled fish is mated with the transparent scaled fish with the result that most of the transparent scaled offspring are heterozygous. A third explanation is that during the classification and counting of the fishes of the F_4 generation containing the three different kinds of fishes I noticed that when the number of fishes in a basin amounted to several hundred with a keen competition of the fishes in the struggle for food, the transparent homozygous fish were always smaller than the other two classes of fish. The common practice of the goldfish breeder is to make a selection of the desirable fishes to rear when the young fishes reach the size of about 20 to 30 mm. At this time the larger fishes are selected to be grown for sale or for breeding and the smaller fishes are discarded or disposed of in other ways. If the transparent fishes are allowed to grow along with other fishes, it is probable that the other classes of fish will be selected in greater number and the transparent homozygous fish will be mostly discarded and only a small portion selected and grown for sale or for breeding. Probably, it is due to all the above mentioned reasons that the homozygous transparent fishes are rare and difficult to obtain from the common goldfish dealer and the existence of this class of fish has not yet been recorded in the goldfish books of any language.

SOMATIC MUTATIONS

In the summer of 1926 I found that among the offspring of the matings between the normal scaled females and males there were six fishes which had a portion of their scales becoming transparent. But these fishes were different from the transparent scaled fishes which I studied. In the ordinary transparent scaled fishes the homozygous forms are nearly completely transparent and without chromatophores and the heterozygous fishes have the characteristic mottled pattern while in the case of the exceptional transparent scaled fishes produced from the mating of two normal scaled fishes the reflecting tissue is only partially reduced and yet the chromatophores do not form the characteristic mottled pattern. These exceptional transparent scaled fishes were of the ordinary red or white color like ordinary normal scaled fishes.

In the spring of 1927 I made two matings of the exceptional transparent scaled fishes with normal scaled fishes. These two matings (E3, E5) produced only normal scaled offspring. In the same breeding season I made one mating (1-9) of the exceptional transparent females and the exceptional males. The offspring obtained from this mating were also all normal scaled as the other normal scaled breeds of goldfish. This result proved that these exceptional transparent scaled fishes were genotypically with the TT genes but phenotypically with some transparent scales. Probably the formation of the transparent scales on the body of such fishes might be due to a somatic mutation of one of the gene T into a gene T' but the germplasm was not affected.

Among the forty fishes which I used to distinguish the homozygous from the heterozygous fish by means of the progeny tests I found one exceptional female fish (B11) which had one side of its body like an ordinary transparent homozygous fish but another side of its body like an ordinary heterozygous mottled fish with a non-transparent operculum and three normal scales, a thing which has never been found in the ordinary homozygous transparent fish. The mating of this fish with a normal scaled male fish produced offspring all belonging to the heterozygous mottled class thus proving that the fish was homozygous for the two $T'T'$ genes. The fact that one side of the body showed the heterozygous condition might be explained by a somatic mutation of one of the T' genes into a T gene during the early stage of development of the fish so that one side of the body was affected.

If the explanations of the above exceptional cases be true we might say that the gene T may mutate to gene T' in the somatic cells and the reverse mutation is also possible.

REVIEW OF LITERATURE AND COMPARISON WITH OTHER CASES

So far as I know there is only one paper on the inheritance of the characters of goldfish. This is the paper of HANCE (1922). In this paper HANCE touched upon the inheritance of colors, telescopic eyes, and the paired caudal fins and concluded, without reporting any data, that these characters were Mendelian. Although there was a picture of the mottled fish in his paper he seemed not to have worked or noticed the simple inheritance of this character. In my breeding work I found the inheritance of the color, telescopic eyes, and paired caudal fin, although with segregations in the F_2 generation, was very complicated whereas the inheritance of the mottled pattern was very simple and followed Mendel's law with the observed ratio very close to expectation. Probably this is the first case of Mendelian inheritance observed in the ordinary goldfish.

The inheritance of the transparency and mottling is a peculiar case of Mendelian inheritance in several respects. The heterozygous condition is a new type distinct from the two homozygous types. The new heterozygous character is a mosaic of the two homozygous types in regard to the presence and absence of reflecting tissue and chromatophores. The genes produce manifold effects on the reflecting tissue, melanophores, and xanthophores. A variegated condition is produced by the contrasting allelomorphic genes. Expression of the genes is somewhat variable. The combination of the above cited peculiarities in a single case is very rare. I found no parallel case in the genetics of animals or plants.

The case of the transparency and mottling is similar to that of the blue Andalusian fowl as reported by BATESON and PUNNET (BATESON 1911). In each case there is a distinct heterozygous type different from the two homozygous types the ordinary distinction of dominance and recessiveness being lacking. The heterozygous type in both cases is a sort of mosaic of the two homozygous types. In the goldfish the heterozygous type is mosaic in regard to the presence or absence of reflecting tissue and chromatophores. In the fowl, the heterozygous type is mosaic in regard to presence and absence of black pigment. However these two cases are not entirely similar since the genes of the goldfish have manifold effects while those of the fowl have only a single visible effect.

The case of transparency and mottling is more similar to that of the roan in the shorthorn cow (BATESON 1911). Here the red color in crossing with the white cow produces offspring with both red and white spots or roan just as the crossing of fishes with reflecting tissue distributed uniformly on the body with fishes lacking the reflecting tissue produces offspring with variegation in the presence or absence of the reflecting tissue. These two cases are also similar in the mosaic expression of the two homozygous types. However they are dissimilar in the presence of the manifold effects in the case of goldfish and the absence of it in the case of cattle.

SUMMARY

1. A pair of Mendelian factors has been definitely found in the goldfish. I propose to call the new character "transparent." This pair of factors is not sex-linked.

2. The homozygous condition of the transparent factor has the following manifold effects: the reflecting tissue is nearly lacking so that the red gills are visible through the operculum, the black retinal pigment is visible through the iris, and the white testis or yellow eggs are visible through the body walls. In such a fish the melanophores are restricted to a few black dots aggregated near the junction between the caudal peduncle and the caudal fin. The xanthophores are generally completely absent. The normal allelomorph of transparent when homozygous causes the production of the reflecting tissue and the two kinds of chromatophores to cover the whole body.

3. The heterozygous condition causes the production of the type of goldfish called "calico" in the United States and "shubunkin" in Japan. This heterozygous form is a mosaic of the two forms of homozygous fish in regard to the reflecting tissue, but the combined action of the two kinds of genes affects the chromatophores in a peculiar way so that a characteristic mottled pattern is produced.

4. In a strict sense there is no dominance and recessiveness between the members of this allelomorphic pair. But, according to the idea of MORGAN, the transparent condition might be said to be dominant and the normal condition to be recessive because the heterozygous mottled condition is sharply distinct from the normal form but is somewhat overlapping with the homozygous transparent condition especially when the fish are smaller than 20 mm in size.

5. Some cases of somatic mutation of the normal gene into the transparent gene have been observed and a case of somatic mutation of the transparent gene into the normal gene has also been observed.

6. Wild goldfishes have been successfully crossed with different kinds of fancy goldfish with the production of fertile offspring.

The above cited investigation was undertaken in the Biological Laboratory of the SCIENCE SOCIETY OF CHINA while I was occupying a position in the Department of Zoology in the College of Agriculture of the NATIONAL

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